Atomic Force Microscopy imaging of ALH84001 fragments.

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Since the announcement by Dr. D. McKay in August last year of the possible existence of primitive fossilised organisms within carbonate globules found on ALH84001, a known Mars meteorite, the scientific community has been divided. The arguments supporting and disclaiming the evidence gathered by Dr. McKay and his colleagues have raged loudly and publicly. Images of structures labelled as possible microfossils have been presented as part of the evidence for primitive life (1). One of the major criticisms of these structures is that they may be artefacts produced during sample preparation required for Scanning Electron Microscopy (SEM). In a further effort to image these structures at high resolution and without sample pre-treatment, fragments of ALH84001 were examined using Atomic Force Microscopy (AFM). AFM is a form of Scanning Probe Microscopy (SPM) which has made imaging at atomic resolution a routine procedure (2,3). In the area of biological imaging, the high resolution and three dimensional capability of SPM, and in particular AFM has been exploited to image a range of proteins (2), nucleic acids (4) and cells (5). AFM is a technique which relies on the interaction of a Si₃N₄ tip attached to the end of a flexible cantilever, with the sample surface. The sample is mounted onto a scanner which incorporates piezoceramics which move in an X-Y raster. The cantilever tip stays 'in contact' with the sample and as the sample moves in the X-Y plane, changes in height of the features on the surface cause the cantilever to be

deflected. This deflection is measured by a laser beam reflected off the back of the cantilever onto a split photodetector (Figure 1). This information is then used to generate a 3-dimensional image of the surface, thereby allowing quantitative measurements of surface topography.

After initial imaging of <u>uncoated</u> fragments using Environmental SEM (ESEM), fitted with Energy Dispersive X-Ray Analysis (EDX), samples were imaged with the AFM. The combination of these techniques has allowed the location and imaging of both the inner surface of the carbonate globules and the iron and magnesium rich rims which surround the globules.

Samples of terrestrial bacterial fossils have been obtained and imaged using SEM and will shortly be imaged employing AFM. Furthermore samples of gold and gold/palladium coated silicon and mica wafers have been imaged using the AFM in an attempt to identify any sources of artifact which may be present on the NASA images of ALH84001.

The results of this preliminary work are encouraging; it has been demonstrated that the AFM can image fragments of ALH84001 at nanometer resolutions. EDX mapping of the fragment surface allowed the AFM to image the different phases of the carbonate globules down to nanometer resolutions. The surface features in these phases appears to differ. In the FeS rich regions there are a considerable number of rounded extrusions from the surface measuring in the range of 100-200 nm in

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diameter (Figure 1). Within the carbonate globules there are no such rounded protrusions and the surface possesses a nanometer scale roughness over all the observed features (Figure 2.). Segmented structures, approximately 500 nm in length have been observed, however at this time only on a single sample, although this may change in the coming months as larger areas of other fragments are explored. The results gained thus far indicate that the structures observed by NASA are not due to the gold/palladium coating used in SEM preparation. Whether these structures are the fossilised remains of a Martian biofilm has yet to be elucidated. However, at the time of writing this abstract, the highest scan ranges utilised have been in the 400 - 500 nm range, higher magnification images will be compiled of the structures which resemble those observed by NASA. These images will then be compared with images of both the base rock and terrestrial bacterial fossils in an attempt to reveal the nature of the structures labeled as microfossils.

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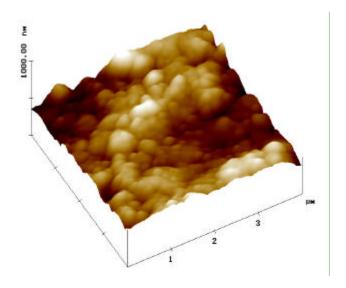
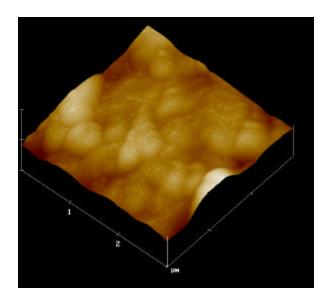


Figure 1. AFM image of iron rich rim of a carbonate globule. Note the rounded protrusions approximately 200nm in diameter.



<u>Figure 2.</u> AFM image within the carbonate rich area of a globule.